
PROTEIN AND CARBOHYDRATE COMPOSITION OF LUCERNE GROWN IN CANTERBURY

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SUMMARY

Carbohydrate fractions and total nitrogen were measured in leaves and stems of Wairau lucerne grown in Canterbury. Leaves had lower levels of total soluble carbohydrate (3-5%), cellulose (8-10%) and lignin (2-4%), and higher levels of total N (3.5-5%) than stems (total soluble carbohydrates 10-22%, cellulose 25-35%, lignin 4-11% and total N 1.5-4%). During the season, leaves did not change much in composition, whereas stems gradually increased in cellulose and lignin. Winter-growing lucerne stems contained high total N (up to 4%), low, cellulose (13%) and lignin (4%). Irrigation increased soluble carbohydrate in stems, but not leaves in immature lucerne. Defoliation of lucerne led to a fall in soluble carbohydrate in the standing stems. About 50% of the lucerne total N was easily extracted from the plant as protein. On a kg/ha basis, a yield of 15, 232 kg of DM (non-irrigated) contained 480 kg of total N, 1,385 kg total soluble carbohydrate and 2,563 kg of cellulose. Over a 70-day summer period, a protein yield of over a metric ton/ha was extracted from irrigated lucerne.

INTRODUCTION

THE NATURE and levels of various constituents in plants grazed by ruminants are of interest not only because of their functions in the plants, but also because of the way they are digested by the ruminant. They can be of further interest in attempts to by-pass the ruminant by preparing from pasture food products for direct human consumption. This paper will consider the composition of lucerne grown near Lincoln, Canterbury, so far as carbohydrates and proteins affect these three aspects.

EXPERIMENTAL

Analytical results are from pure stands of Wairau lucerne on Wakanui silt loam at Lincoln. Plots were sampled or

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TABLE 1: CARBOHYDRATE AND NITROGEN CONTENT OF LUCERNE LEAVES AND STEMS
(% on DM basis)

LEAVES								
Date.	Harvest	Growth Stage*	Total N	Soluble Carbo- hydrate	Pectin	Hemi- cellulose	Cellu- lose	Lignin
28.11.68	1	4	3.94	5.7	6.5	7.3	8.8	2.5
23.12.68	2	1	5.34	7.2	4.4	6.8	5.6	3.1
8. 1.69		2	4.42	4.8	6.1	8.7	6.1	3.9
20. 1.69		3	4.02	7.1	3.7	4.0	9.0	4.3
29. 1.69		4	3.78	8.1	4.2	7.3	11.0	4.3
24. 3.69	3	4	3.52	9.7	4.6	4.7	6.2	3.8
28. 4.69	4	1	4.77	7.4	4.0	5.3	6.4	3.6
27. 5.69		2	4.72	6.8	6.0	5.5	7.5	4.4
15. 6.69		3	4.03	12.7	5.6	5.2	7.6	3.8
8. 9.69	5	3	5.52	4.6	2.7	6.0	7.5	—
11.11.69	6	4	4.08	7.0	4.2	6.7	8.1	2.5
2.12.69	7 dry	3	5.06	6.6	3.5	5.0	7.9	3.3
	Irrig.	3	5.50	7.4	5.6	4.2	8.1	3.5
12.12.69	dry	4	4.58	7.6	6.2	7.8	8.6	3.5
	Irrig.	4	5.27	6.5	5.6	5.6	9.2	3.4
20. 1.70	8 dry	4	4.54	6.5	8.2	5.0	10.5	8.9
	Irrig.	4	5.00	4.4	9.0	4.0	12.0	9.5
S.E.			0.11	1.3	0.3	1.3	1.4	0.2

STEMS								
28.11.68	1	4	1.84	13.2	4.9	12.1	30.1	8.5
23.12.68	2	1	3.28	13.5	4.8	6.8	13.9	3.6
8. 1.69		2	1.64	10.2	4.5	8.3	25.3	7.6
20. 1. 69		3	1.37	10.1	3.6	8.2	34.2	11.0
29. 1.69		4	1.42	8.2	4.2	9.8	35.0	10.5
24. 3.69	3	4	1.34	13.1	4.3	9.9	23.5	9.6
28. 4.69	4	1	3.41	17.5	4.8	7.6	15.7	3.8
27. 5.69		2	3.49	18.0	9.3	7.5	13.6	4.3
15. 6.69		3	4.03	22.5	7.2	8.1	13.5	5.7
8. 9.69	5	3	4.21	12.2	2.7	5.3	12.0	—
11.11.69	6	4	1.96	12.8	4.5	11.9	23.5	7.3
2.12.69	7 dry	3	2.62	9.2	4.0	6.6	17.7	4.5
	Irrig.	3	3.98	15.2	7.2	5.4	15.4	4.0
12.12.69	dry	4	2.10	8.6	5.2	9.8	25.8	8.5
	Irrig.	4	2.64	12.8	6.7	8.8	26.0	6.9
20. 1.70	8 dry	4	1.44	9.0	6.8	9.5	33.4	11.5
	Irrig.	4	1.78	7.5	7.8	9.4	33.1	10.6
S.E.			0.14	0.8	0.9	1.2	0.5	0.5

* Stage 1, 10-15 in. high; 2, 15-20 in. high; 3, 20-35 in.; 4, early flowering.

harvested on the dates shown in Table 1. Samples were taken at intermediate stages of growth (stages 1, 2 and 3, Table 1) and finally at early flowering (stage 4, Table 1). Total lucerne was harvested at this later stage except in winter and early spring, when it was harvested at the equivalent of stage 3 when older leaves had commenced to fall. Samples for carbohydrate analyses were taken, between 1 and 2 p.m. in bright conditions, from two pairs of plots, separated into leaves (including buds and flowers) and stems and freeze-dried. Dry matter yields and samples for nitrogen analyses were taken from 2m² quadrats in each of four pairs of plots, dissected as above and oven dried. Irrigation was applied where indicated, at approximately 20% available soil moisture, to one half of the plots. Carbohydrates and lignin were measured using essentially the analyses scheme of Bailey (1967). Total N was measured by the standard Technichon autoanalyser system after Kjeldahl digestion. True protein extractions were performed on material from dry and irrigated plots (7/4 and 8/4, Table 1) using standard pulping and pressing equipment (Davys and Pirie, 1969; Davys *et al.*, 1969). True protein was measured in the press juice by precipitation with 5% w/v trichloroacetic acid and non-protein nitrogen was measured in the supernatant.

Supplementary studies of the effect of defoliation on stem soluble carbohydrates involved Wairau lucerne on Templeton, and Kaiapoi soils during November 1969.

RESULTS AND DISCUSSION

PROTEIN COMPOSITION OF LUCERNE

Total nitrogen contents of leaves and stems are shown in Table 1. In all of this work total nitrogen is taken as an indication of protein and is an overstatement because of the presence of non-protein N to the extent of approximately 10%.

Differences were significant between seasonal harvests in nitrogen levels in both leaves and stems. In all harvests except in winter and early spring, N contents of stems were very much lower than those of leaves. The interaction between plant parts and season was significant at the 1% level. Sequential samples up to the summer harvest of 1968-9 (harvest 2) and up to the winter harvest (harvest 4) show much more marked decline in N level with advancing growth in summer than with the much greater passage of time in winter, especially in stems,

These changes must, however, also be considered in terms of total yield/ha when it can be seen (Table 2) that N yield is highest at the most advanced growth stage. Irrigation significantly (at the 1% level) increased the levels of total N in both leaves and stems.

Because of differences in sampling and growth conditions the many lucerne N and protein analyses scattered through the literature are not always comparable with present results. High levels of N in leaves (4 to 4.5%) and low levels in stems (1.5%) have been reported by Heinrichs *et al.* (1969) from Canada and by Couchman (1960) from Australia. Many authors have recorded high levels of protein (up to 30%) in total lucerne (e.g., Van Riper and Smith, 1959; Dent, 1959).

CARBOHYDRATE COMPOSITION OF LUCERNE

Results of the carbohydrate analyses of lucerne leaves and stems are given in Table 1. Total soluble carbohydrate (extracted with boiling water and ranging from simple sugars such as glucose and sucrose to polysaccharide such as starch) is the photosynthate or energy-supplying carbohydrate available for structural carbohydrate or protein synthesis or for translocation to root reserves. Its levels will fluctuate greatly according to weather and growth conditions. Pectin appears to function as a kind of inter-cellular cement while hemicellulose and lignin form an amorphous cell-wall matrix around the more highly organized and fibrous cellulose. These various carbohydrates are present in all plant tissues but legumes differ from pasture grasses in that they are richer in pectin.

Present results show clear differences between leaves and stems, stems being higher in soluble carbohydrates, cellulose and lignin. Diurnal fluctuations in soluble carbohydrates in lucerne leaves and stems were examined in samples taken at intervals during 24 hr in warm, bright weather in November 1969. The levels in apical leaves fell from a day maximum of 10 to 11% at 3 p.m. to a night minimum of 5.6% at 5 a.m. In the same period, stem levels fell from 15.9% (3 p.m.) to 13.8% (5 a.m.). Sequential sampling in summer growth indicates that leaves do not change much in composition, whereas in stems there is a decline in soluble carbohydrates and an increase in cellulose and lignin. These changes in stem composition are not paralleled in winter growth. Comparisons of results from successive harvests at advanced growth stages (3, 4) suggest that the main seasonal effects are lower levels of

cellulose and lignin in the stems of the slower grown winter and early spring harvests when flowering was not evident. There is little evidence of marked fluctuations in structural carbohydrates among the harvests of the main production period. Irrigation apparently can have a marked effect on the level of soluble carbohydrate in stems but not in leaves. While this effect may be seasonal, it could be confined to less mature stages of growth as it was absent from the 8/4 sample (Table 1) with more advanced flowering than the 7/3 or 7/4 samplings. The increase possibly reflects a more active photosynthesis in irrigated plants and hence more photosynthate for translocation to the roots.

In lucerne grown in Scotland, Hirst et al. (1959) found structural carbohydrate levels similar to the present ones with the same type of leaf-to-stem differences. Their lucerne contained, however, much lower levels of soluble carbohydrates in the stems. While Van Riper and Smith (1959) in Wisconsin record crude fibre values **similar** to our cellulose levels, Heinrichs et al. (1969) in Canada and Lagowski et al. (1958) in Michigan found much higher levels of structural carbohydrates (up to 50% cellulose in stems). The latter authors also found only very low (3-6%) soluble carbohydrate levels in stems as did Nishikawa (1967) in Japan. Higher levels of soluble carbohydrate in whole lucerne plants have been recorded.-e.g., Burns et al. (1964), 10 to 13%, and Melvin (1965), 6 to 12%. These differences do suggest that the high levels of soluble carbohydrate encountered in Canterbury Wairau lucerne (up to 22-24%) may be exceptional.

THE IMPORTANCE OF CARBOHYDRATES AND PROTEIN OF LUCERNE TO THE GRAZING ANIMAL

Individual plant carbohydrates are digested at **different** rates and to different extents by ruminants. Soluble carbohydrates and probably pectin which are rapidly and completely digested represent readily available energy precursors. Structural carbohydrates are more slowly digested with much (up to 30% of the cellulose) escaping **digestion** probably because of **lignification**. These latter carbohydrates represent less readily available energy. The important role of the ruminant is that of digesting these plant structural carbohydrates which have been locked in cell walls, and which are unavailable to man. In evolving the **rumen** system to utilize this range of plant carbohydrates, the ruminant, however, has sacrificed efficiency in utilizing protein. The **rumen** system of hydrolysing

ingested protein to ammonia and amino acids which are incorporated into microbial protein for subsequent digestion and absorption in the lower digestive tract leads, on high protein diets, to direct absorption of much of the ammonia and hence to its urinary excretion as urea.

The analyses of Table 1 show that lucerne stems are richer than leaves in both forms of energy although in the more mature stems lignification will lower the cellulose digestibility. Hence in summer growth maturing stems will probably be the least digestible part of the plant. Lucerne leaves in particular are an example, so far as **herbage** is concerned, of a high protein diet which the grazing animal will use inefficiently.

Published opinions differ on the extent to which lucerne may be regarded as a high quality ruminant feed (Campbell, 1967). Present analyses indicate that leafy tops (apices plus top stem), immature lucerne or cool season growth should be a high quality feed which is in agreement with the liveweight gains reported by McLean *et al.* (1967) for sheep feeding selectively on lucerne.

As grazing sheep preferentially remove lucerne apices and leaves (O'Connor, 1970) the influence of defoliation on the total soluble carbohydrate in stems was examined in November 1969 at two sites. Pre-flowering stage 3 lucerne was left intact, stripped of apex and uppermost leaves or stripped of all leaves and the upper portion of

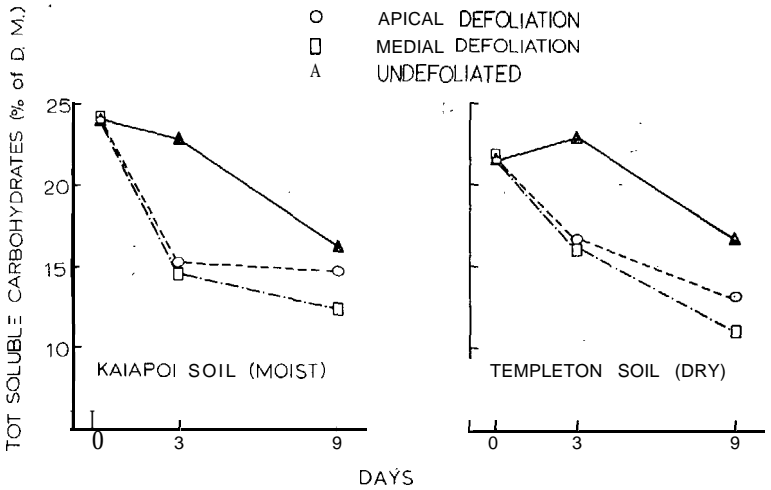


Fig. 1: Total soluble carbohydrates in lucerne stems during November 1969 with and without defoliation.

stem. Although the soluble carbohydrate in intact stems declined over a nine-day period, its decline was more rapid in defoliated stems. (Fig. 1) and appeared to be greater in the more completely defoliated stems.

Although preferential grazing of apices in spring leaves stems which will be fairly high in soluble carbohydrate, these stems will decline in quality through loss of soluble carbohydrate if grazing pressure is low. The lower levels of readily available energy precursors and protein in mature summer lucerne stems may account for the reluctance of sheep to eat these stems, especially if the preferential removal of leaves by the grazing animal leads to a further fall in the soluble carbohydrate.

Yields of carbohydrate and protein are presented on a kg/ha basis in Table 2. These results further emphasize the extent to which the bulk of the carbohydrate is contained in the stems, except in immature and winter growth.

DIRECT EXTRACTION OF PROTEIN FROM LUCERNE

Lucerne is the crop chosen for study in a DSIR investigation into the extraction of leaf protein for direct consumption by humans or other monogastric animals. Both dry matter and total nitrogen yields (Table 2) indicate the potential of lucerne as such a source of protein. About 480 kg/ha of nitrogen were harvested in 15,232 kg/ha of dry matter in the five harvests between September 1968 and September 1969 (Harvests 1-5). Considerably higher yields of **herbage** have been obtained at Lincoln (Vartha and O'Connor, 1968; O'Connor *et al.*, 1968) under better rainfall regimes than those experienced in 1968-9. The distribution of total nitrogen between stems and leaves during 8 months of the year favours leaves alone as a source of protein for extraction. Extraction studies so far have been confined to the whole plant. The early summer harvests (7/4) yielded 54% and 51% of their total-N as extractable true protein for the dry and the irrigated forages respectively. The midsummer harvest 8/4 gave similar percentage yields of true protein-N (44% and 46%), for dry and irrigated conditions. So far we have no evidence that irrigation has materially affected the net. extraction rate for protein. Nevertheless, irrigation at least doubled the total yield of extractable protein; (e.g., early summer harvest 43.1 to 103.5 kg/ha protein-N, and midsummer harvest 32.4 to 57.8 kg/ha protein-N). These yields represent, over 70 days' growth, an extracted protein yield of 476 kg/ha or 6.8 kg/ha/day from **dryland** and 1,029 kg/ha or 14.7 kg/ha/day from irrigated lucerne.

TABLE 2: YIELDS OF LUCERNE DRY MATTER, NITROGEN AND CARBOHYDRATES
(kg/ha)

Harvest	Growth Stage*	Leaves				Stems			
		D.M.	N	Total Soluble Carbo- hydrate	Cellulose	D . M .	N	Total Soluble Carbo- hydrate	Cellulose
2	4	2,694	190	148	229	3,323	58	438	1,000
				67	52	519	17	70	72
	2	1,831	81	88	112	1,526	25	156	522
	3	2,178	87	155	196	2,212	30	234	756
	4	2,514	95	204	276	1,962	28	161	686
3	4	825	29	80	51	492	7	64	116
4	1	477	23	35	30	142	5	25	22
	2	568	27	39	43	136	4	24	19
	3	1,021	41	128	77	301	10	68	41
5		1,465	81	67	110	734	30	90	88
1968-9	total yield	8,430	346	632	743	6,812	134	753	1,931
6	4	1,982	88	141	160	2,492	35	327	568
7	3 dry	674	34	51	51	278	7	26	49
	3 irrig.	1,185	65	88	96	486	19	74	75
7	4 dry	1,314	60	100	113	936	19	80	241
	4 irrig.	2,624	138	171	241	2,573	68	328	668
8	4 dry	1,253	57	81	132	1,113	16	102	377
	irrig.	1,693	84	74	203	2,236	40	177	740

* Dates and growth stages in Table 1.

The extraction process envisages the utilization of the residues for ruminant feeding. The process produces a fibrous bagasse, principally cellulose, hemicellulose, pectin and lignin with some soluble carbohydrate, protein, non-protein-N (NPN) and minerals and a whey containing mainly soluble carbohydrate, NPN and minerals. NPN, as a percentage of total N, has generally been about 10% for summer-grown material pulped for extraction, most of which is released in the juice. Jasiorowski (1960) found high arginine contents (600 mg% on an air-dry basis) in lucerne hays and suggested this had harmful effects on the nitrogen balance in ruminants but we found such low levels of arginine that we would not expect any comparable harmful effects from the feeding of fibrous residues. While the bagasse may be fed direct to ruminants, the whey is a disposal problem and potential loss of valuable nutrients. This loss would be minimized if protein was extracted from leaves only when most of the soluble carbohydrate would remain with the stems which could be fed separately.

CONCLUSIONS

Canterbury lucerne is a crop yielding large amounts of readily available carbohydrate energy as well as fibre, and large amounts of protein. Efficient utilization of this energy could well incorporate a siphoning off of large amounts of the protein for direct human consumption.

ACKNOWLEDGEMENTS

Acknowledgements are due to 'the technical staff of Grasslands Division, DSIR, Lincoln, for collecting and preparing samples and to members of the analytical groups, Applied Biochemistry Division, DSIR, Palmerston North and Lincoln, for help in the analyses.

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DISCUSSION

Asked by Reid (U.S.A.) if digestibilities had been measured in relation to herbage intake by animals, Bailey replied that they had not since the set-up did not provide for animal work. Marten (U.S.A.) asked if there had been any differences in protein and carbohydrate contents which could be caused by temperature within and between seasons. Bailey said there was evidence that the leaves were higher in soluble carbohydrate in winter. This could be due to the fact that winter lucerne did not reach the mature stage. Langer asked if there was any information on the annual total nitrogen yield of lucerne which could be compared with annual total nitrogen production by white clover in the same environment. Bailey replied that the figure for lucerne was 480 kg/Ha but he had no comparable figures for white clover. Evans asked what the amino acid composition of protein was in terms of its suitability as a source of human food. Allison replied that it was better than the FAO standard for lysine in particular. It was marginal only in thiamine and cystine. He suggested it would be as good as, if not better than, skim milk. Mitchell inquired if crops other than soybean and lucerne were available, to which Bailey replied that lucerne was being worked on in Canterbury. O'Connor found it difficult to visualize how the animal consumption pattern could be got round to fit the human pattern without going to expensive means of conversion or conservation. It would be better to have a complementary system to make use of the surplus as human food. Butler commented that Stahlman had investigated the possibility of extraction from a wide range of plant wastes. In Britain, also, many sources of extractable protein had been investigated. Wedin (U.S.A.) reported that, in Iowa, engineers had designed a leaf-stripping machine to remove most leaves but to leave sufficient for recovery growth. Some manufacturers were investigating the possibility of using these leaves in the production of edible protein. O'Connor suggested that perhaps engineers could devise a machine to remove a proportion of leaves so that sufficient remained for animal requirements.
